High-throughput epitope identification for snakebite antivenom

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High-throughput epitope identification for snakebite antivenom

Mikael Engmark¹, Federico De Masi¹, Andreas Hougaard Laustsen², José María Gutiérrez³, Bruno Lomonte³, Mikael Rørdam Andersen¹, and Ole Lund¹

Introduction

Insight into the epitopic recognition pattern for polyclonal antivenoms is a strong tool for accurate prediction of antivenom cross-reactivity and provides a basis for design of novel antivenoms. In this work, a high-throughput approach was applied to characterize linear epitopes in 966 individual toxins from pit vipers (Crotalidae) using the ICP Crotalidae antivenom. Due to an abundance of snake venom metalloproteases and phospholipase A₂ in the venom used for production of the investigated antivenom, this study focuses on these toxin families.

Objectives

- Identify epitopes in toxins used in immunization
- Characterize tolerated amino acid substitutions in identified epitopes
- Predict cross-reactivity of antivenom

Epitopes locate to surface regions

To identify epitopes observed single specific signal intensities were mapped back to the amino acid sequence of each pit viper toxin. Using two or more overlapping 15-mer peptides with median signals above 20 AU, epitope core sequences were localized and subsequently mapped to crystal structures or homology models. As examples, P-I metalloproteinase and lys49-phospholipase A₂ from Bothrops asper (venom used in antivenom production) are presented.

Immunization mixture¹

Studying linear epitopes using peptide microarrays

Mean AU overlapping peptides

Effect on cross-recognition

In silico generation of peptide library

Antibody binding and detection

The peptide microarray experiments translate to the snakebite antivenom. Due to an abundance of snake venom metalloproteases and phospholipase A₂ in the venom used for production of the investigated antivenom, this study focuses on these toxin families.

References


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